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THETA ACTIVITY AND
RUNNING SPEED IN THE RAT**

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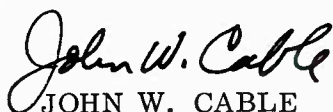
RECORD SET

Research was conducted according to the principles enunciated in the
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RELATIONSHIP BETWEEN HIPPOCAMPAL THETA ACTIVITY
AND RUNNING SPEED IN THE RAT

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FOREWORD
(Nontechnical summary)

The hypothesis that the hippocampal theta rhythm (6-8 Hz) is related to movement was tested by recording bioelectric activity from the hippocampus and cortex of rats while they were running at various speeds on a treadmill. The results indicate that the theta rhythm is directly related to locomotor activity and that EEG frequencies outside the theta band do not change as a function of running speed.

ABSTRACT

To test the hypothesis that hippocampal theta activity is related to movement, chronic EEG recording electrodes were implanted in the dorsal hippocampus and cortex of six rats. The rats were then forced to run at varying speeds on a treadmill while their brain bioelectric activity was being recorded. A close relationship appeared between amount and amplitude of hippocampal theta activity and speed of running. No such relationship was evident in cortical activity. Frequency analysis by means of a Schmitt trigger and electronic filter also demonstrated the development of 6- to 8-Hz (theta) activity in the hippocampus with running but no corresponding enhancement of 10- to 12-Hz (nontheta) activity.

I. INTRODUCTION

A 6- to 8-Hz (theta) rhythm often becomes a prominent feature of the bioelectric activity of the hippocampus in a variety of species. Conjectures as to the conditions under which this theta activity arises and its biological significance include hypotheses that it is involved in learning and memory consolidation,⁴ orienting responses,^{1,5} or movement.^{2,3,7,9-14} The latter hypothesis is especially interesting because movement of the experimental subject has not been controlled in experiments designed to examine other hypotheses concerned with the significance of hippocampal theta activity.^{4,5} Previous work in this laboratory⁹ has shown that hippocampal bioelectric activity becomes synchronized around a 7-Hz peak and its amplitude increases relative to inactive periods in rats trained to increase their activity levels.

To establish whether the particular conditioning procedure or the movement itself led to the induction of theta waves in the hippocampus of trained rats, it was necessary to induce changes in locomotion by means other than operant conditioning based upon positive reinforcement. In the present study, rats are forced to run at varying speeds on a treadmill to determine relationships between speed of movement and changes in hippocampal bioelectric patterns.

II. METHODS AND MATERIALS

Six male Sprague-Dawley rats weighing 250-370 grams were anesthetized with sodium pentobarbital (50 mg/kg) and placed in a Kopf stereotaxic instrument. Twisted pair, Teflon-insulated, stainless steel wire electrodes were implanted in the dorsal hippocampus (coordinates 2.0 mm behind the bregma, 2.0 mm lateral, 4.5 mm below the skull surface) and miniature, gold-plated Poke-Home (Amphenol Company) pins placed bilaterally on occipital cortices.

After allowing 1 week for recovery from surgery, EEG recording leads were attached and the rats placed on the treadmill illustrated in Figure 1. The bioelectric signals were amplified differentially by Tektronix FM122 units with a band-pass setting of 0.2 - 50 Hz and recorded on both a Dynograph paper chart recorder and on a 7-channel Ampex SP-300 tape recorder. A graphic time code was also recorded on both instruments. Later, the signals from both cortex and hippocampus were fed through a Krohn-Hite 3550-R filter set at band-passes of 6-8 Hz or 10-12 Hz. At the 6- to 8-Hz setting, half power points were 5.4 Hz and 9.1 Hz, at the 10- to 12-Hz setting, half power points were 8.5 Hz and 14.3 Hz. The filter output was then passed through a Schmitt trigger set to fire at a predetermined voltage level. The trigger outputs were recorded on a cumulative recorder. The threshold firing level of the Schmitt trigger was different for cortical and hippocampal signals due to differences in signal amplitude but was always the same for a given structure in a given rat.

The rats were initially placed on the treadmill with the motor off, and a baseline (B1) EEG secured. Next, the rat was placed on a Plexiglas sheet suspended above the treadmill belt and the motor turned on. This served as a control for possible artifacts generated by the motor. The Plexiglas was then removed and the rat was forced to run at a variety of speeds ranging from 1 to 22 cm/sec for about 5 minutes at each speed. A second base line (B2) was obtained with the motor off at the end of each day's running session. At least 3 days of running sessions were obtained from each rat. At the conclusion of the experiment the rats were sacrificed, perfused, and the brains sectioned to confirm the location of electrodes. A representative section showing the location of the hippocampal electrode is shown in Figure 2.

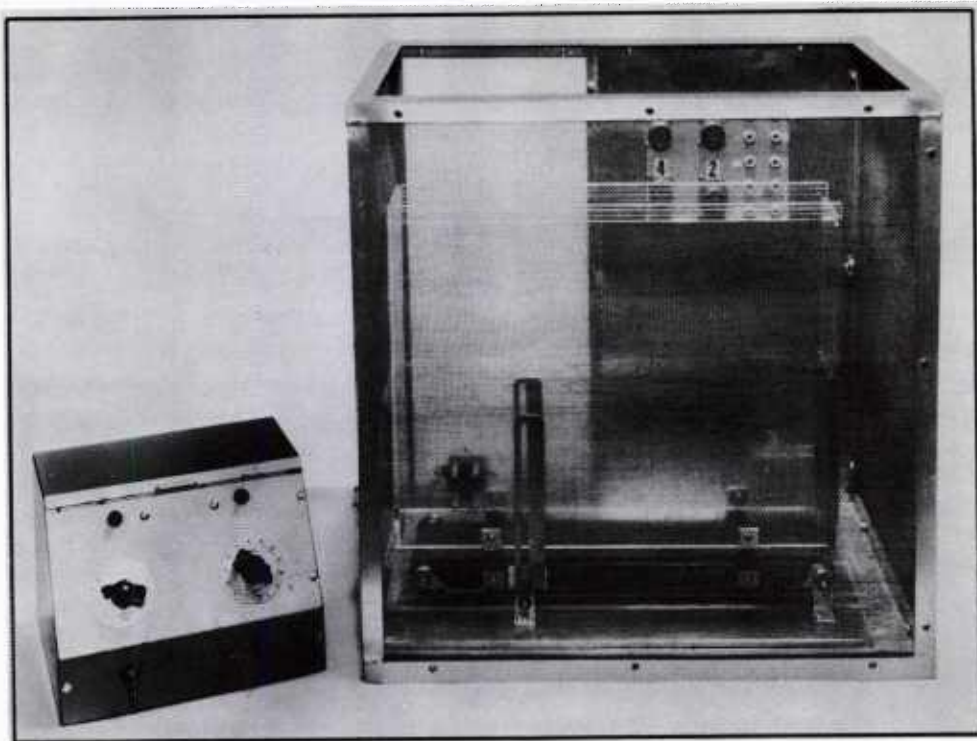


Figure 1. Rat treadmill and control box. The treadmill is placed inside a screened Faraday cage (top not on in this view) with the junction box for EEG recording leads seen at the top back. The motor is located outside the cage in the back and is connected to the treadmill by a plastic, nonconducting shaft.

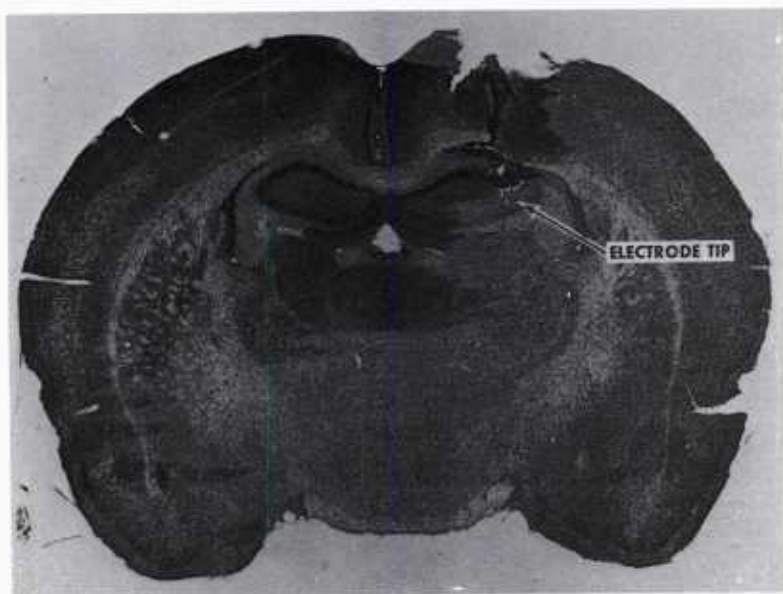


Figure 2.
Representative thionine-stained section showing location of electrode tip in the dorsal hippocampus

III. RESULTS

As soon as the treadmill was turned on, the hippocampal EEG showed marked changes. The amplitude of the EEG increased and the tracing changed from asynchronous activity to activity synchronized at about 7 Hz. The cortex did not show such systematic changes in frequency or amplitude. This phenomenon is clearly illustrated in Figure 3, where a comparison is made of cortical and hippocampal recordings obtained from a rat at rest (B1, B2) and while running at various speeds. In the hippocampal tracing, note that there is little change from base-line to motor conditions. The signals are of variable amplitude and there is little theta present. At a speed of 1 cm/sec, theta activity increases but there is no significant amplitude change. As

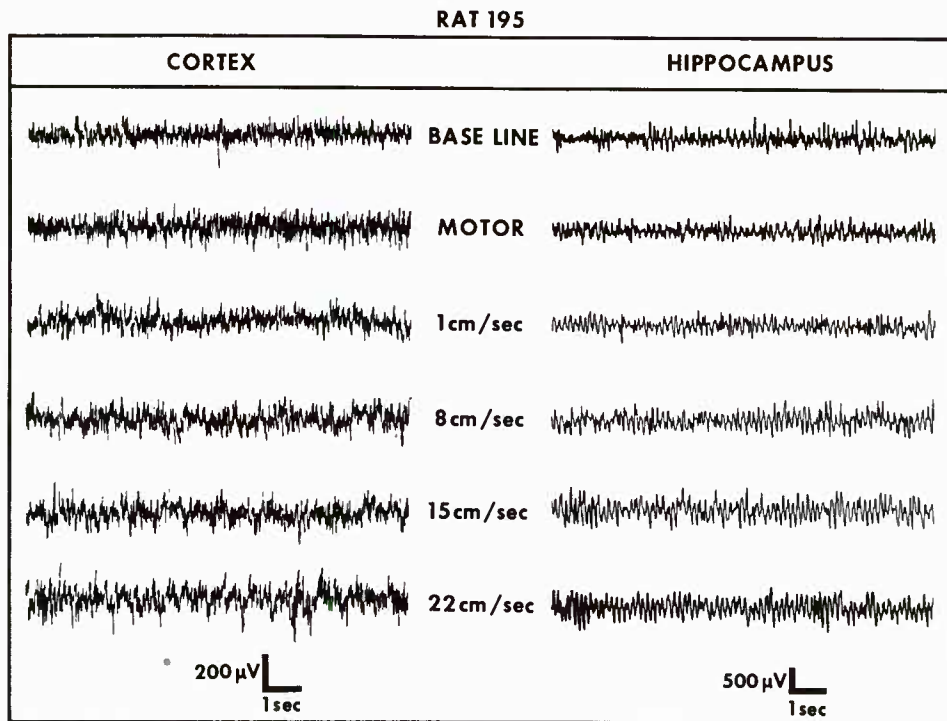
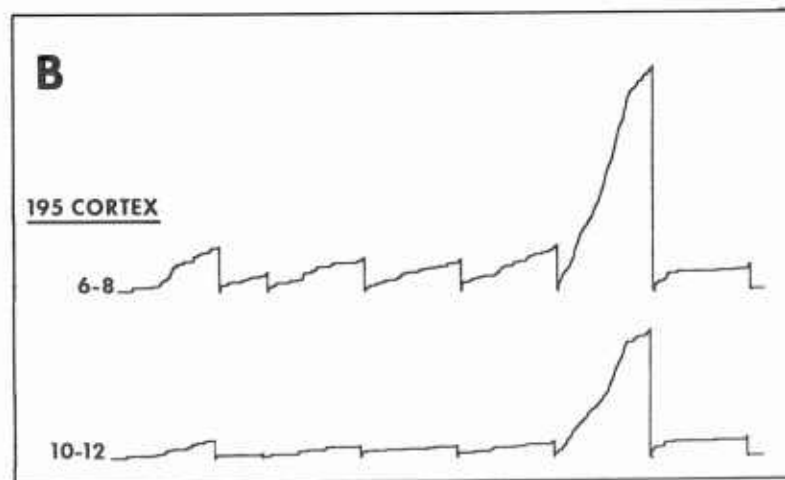
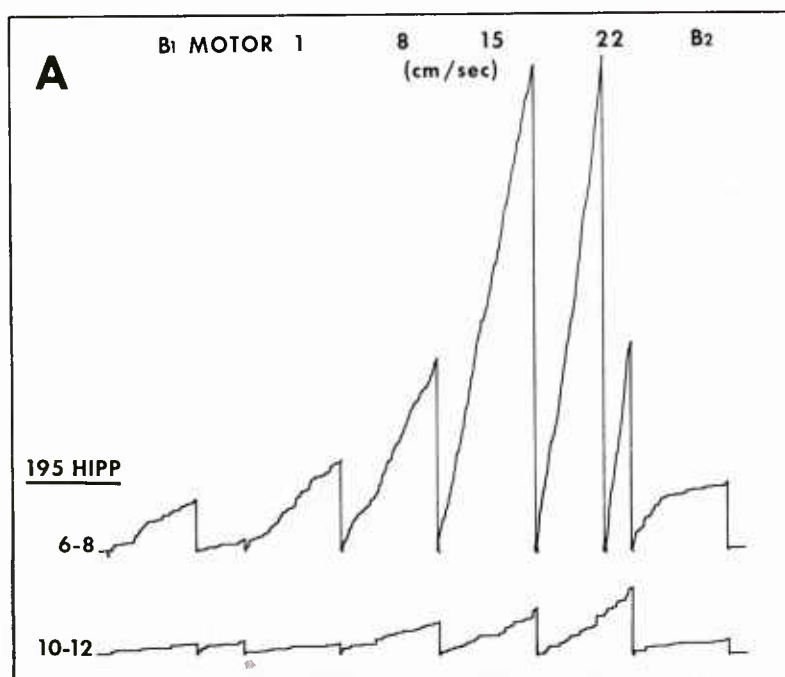


Figure 3. Examples of cortical and hippocampal EEG tracings taken at identical time periods under the indicated conditions. See text for details.

the treadmill speed increases to 8 cm/sec, the amplitude increases and there is greater synchronization of the wave forms at the theta frequencies. Both synchronization and amplitude steadily increase to reach their maximum at 22 cm/sec treadmill speed. In contrast, the cortical tracing shows only a slight amplitude increase at 15 and 22 cm/sec and there is no noticeable synchronization at theta frequencies at any speed.

The cortical and hippocampal recordings shown in Figure 3 were filtered through narrow windows of a Krohn-Hite band-pass filter. The output of the filter varies as a function of the EEG voltage between the given frequency settings. The filtered EEG signals of rats running at different treadmill speeds were then fed into a voltage sensor (Schmitt trigger). Pulses from the trigger actuated a relay in a cumulative recorder which provided a graphic record of changes in output of the filter set at frequency bands within (6-8 Hz) and outside (10-12 Hz) the theta band. The results of this analysis are shown in Figure 4A and B. The conditions B1 and B2 refer to base-line conditions where the animal was sitting quietly. In the "motor" condition the motor was turned on, but the treadmill was disengaged to control for possible electrical artifacts. In Figure 4A, the results of the analysis of the hippocampal EEG are presented. A comparison is made of changes in the 6- to 8-Hz band (theta) and the 10- to 12-Hz band (nontheta) as a function of different experimental conditions. It is quite clear that there is a greater change of activity in the 6- to 8-Hz band than in the 10- to 12-Hz band when the rat is forced to run on the treadmill. Furthermore, the rate of change is proportional to the speed the animal is forced to maintain. This clear-cut relationship is not seen in the analysis of the cortical tracings, Figure 4B, where

no consistent changes are present in either frequency band as a function of running speed.



The systematic change shown in the theta component of the hippocampal tracing was seen in every animal. However, no consistent frequency change was found in the cortex. At the highest speeds, there was an increase in signal amplitude but this was not specific to either frequency band.

While there was a very good correlation of the induction of theta with the onset of movement, hippocampal theta did not stop immediately with cessation of movement (Figure 5). Occasionally, cortical tracings which look identical to hippocampal theta were recorded. Some investigators¹⁵ explained these recordings on the basis of volume conduction from hippocampus. Recordings from animal #181, Figure 5, have a direct bearing on this question. At the top of the figure are tracings A, obtained while the rat is running at a very fast speed. The middle tracings B were taken immediately after cessation of running, and the lower tracings C, 6 minutes after the running ended. Note the differences in synchrony and amplitude of the hippocampal tracings. Theta waves are most prominent in A, and only sporadically present in C. In contrast, theta waves are continuously present in the cortex, and in C where they are less evident in the hippocampus. This means that independent sources for the generation of theta waves are present at these loci.

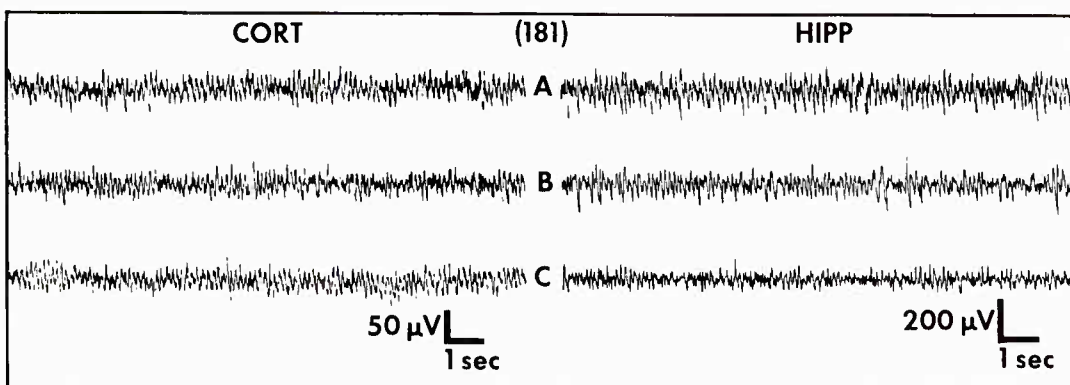


Figure 5. Cortical and hippocampal EEG tracings of a rat running on the treadmill at 18 cm/sec (A); immediately after the cessation of running (B); and 6 minutes after the running ended (C).

IV. DISCUSSION

By demonstrating a relationship between bodily activity and generation of hippocampal theta activity, these data clearly support our previous findings⁹ and those of Vanderwolf and Heron,¹² Vanderwolf,^{10,11} Whishaw,¹³ and Whishaw and Vanderwolf.¹⁴ Furthermore, these results extend the previous findings by showing a nearly linear relationship between amount of hippocampal theta activity and speed of movement.

The results of this study showing that hippocampal theta waves persist for some time after the cessation of running and the findings of Black et al.² indicate that increased muscle tone is not a necessary condition for the occurrence of hippocampal theta waves.

Komisaruk⁸ has shown a correlation between hippocampal theta and rhythmic movements of the vibrissae in the rat. It is possible that while our rats were sitting quietly after a running session they were breathing rapidly, which may explain the persistence of hippocampal theta. Gray⁶ has been able to dissociate such rhythmic vibrissal movements from hippocampal theta activity by means of medial septal nucleus lesions, and hence this explanation is questionable. As indicated in Figure 5, it is possible to dissociate cortical theta waves from hippocampal theta waves indicating that the hippocampus is not necessarily the source of the invasive hippocampal theta waves to the cortex described by Yamaguchi et al.¹⁵

The behavioral procedure used to induce hippocampal theta waves in the rat provides a very simple and reliable method to study the anatomical origin and neurochemical basis of the hippocampal theta waves. Future work will assess the effects of various pharmacological agents on behaviorally induced theta waves. In addition,

anatomical studies involving the destruction of afferent pathways to the hippocampus may provide more information on the generators of the theta rhythm.

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